Association of MAP specific ELISA-responses and productive parameters in 367 Danish dairy farms

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PROCEEDINGS of the 12th ICP

TABLE OF CONTENTS

Opening Ceremony (Jubilee Session)		**2**
Session 1: Pathogenomics and MAP Biology		
	Oral	**4**
	Poster	**16**
Session 2: Diagnostics and Detection		
	Oral	**37**
	Poster	**57**
Session 3: MAP Control Programs		
	Oral	**111**
	Poster	**132**
Session 4: Host Response and Immunology		
	Oral	**163**
	Poster	**176**
Session 5: Genotyping and MAP Diversity		
	Oral	**222**
	Poster	**233**
Session 6: Epidemiology		
	Oral	**251**
	Poster	**265**
Session 7: Public Health and MAP in the Environment		
	Oral	**295**
	Poster	**309**
Author Index		**325**

Abstract P-03.8
ASSOCIATION OF MAP SPECIFIC ELISA-RESPONSES AND PRODUCTIVE PARAMETERS IN 367 DANISH DAIRY FARMS

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Introduction
The impact of *Mycobacterium avium* subsp. *paratuberculosis* (MAP) infection on productive parameters such as milk yield and reproductive features is central in the culling decision on individual cows. This study aimed to compare the test-day milk yield and lactation specific conception probability in 94,064 Holstein cows in 314 dairy herds from the Danish control programme on paratuberculosis (Nielsen, 2007).

Methods
Herds participating in the Danish programme are tested 4 times per year using a commercial MAP specific antibody ELISA (Nielsen et al., 2013), which has been used since 15 October 2008. All data used are from this date and forward. A sample-to-positive ratio >0.2 was considered positive.
The milk yield is recorded either six or eleven times per year depending on the farm. For each of the 314 Holstein herds, these samples were pooled to fit Wilmink lactation curves for parity 1, 2, and 3+ per herd (Wilmink, 1987), leading to a total of 942 lactation curves. These curves represent the expected energy corrected milk yield of the average cow on each farm as a function of days in milk. For each milk yield point, the value relative to the average cow on the farm was then calculated. This relative energy corrected milk yield (rECM) indicated whether a cow at a certain test date had milked as the average cow on the farm (rECM=1) or for example 5% more (rECM=1.05), or 5% less (rECM=0.95). We then tested whether rECM depended on the MAP ELISA score. We also assessed if cows with no positive ELISA test at the test date but with a future positive test would differ in rECM from cows that never tested positive. This was tested using bootstrapping with 10,000 replicates under the null hypothesis that there is no difference in means between groups.

The conception probability was tested for the first three insemination attempts between the first and second lactation period. The cows were divided into two groups: one with animals that had no ELISA positive tests during their lifespan; and one for cows with a positive test result later than the fourth insemination attempt. Thus, no cows testing positive before the four insemination attempts are included. This procedure was used to avoid that a positive test would have caused a farmer to cull the cow, which could potentially change apparent conception probability. The conception probability was then tested in a generalized linear model using the binomial family with a logit link function, with farm and positive/negative ELISA test as fixed effects. Subsequently the test was also carried out using only cows that achieved a 2nd parity.

All data handling, analysis, and plotting were done using the free open source statistical software R (r-project.org). All data were extracted from the Danish Cattle Database (Knowledge Centre for Agriculture, Aarhus, Denmark, vfl.dk) for the use in the ‘iCull’ project (icull.dk).

Results

Test-day milk yield was negatively correlated with increasing milk ELISA value (Figure 1). The figure shows that an increase in MAP S/P ratio is correlated with a decrease in milk production relative to the average cow on each farm. At a MAP S/P-value of 1, the rECM was 0.94, while it was 0.78 at the MAP ELISA value of 5. Differences in conception probabilities are illustrated in Figure 2.

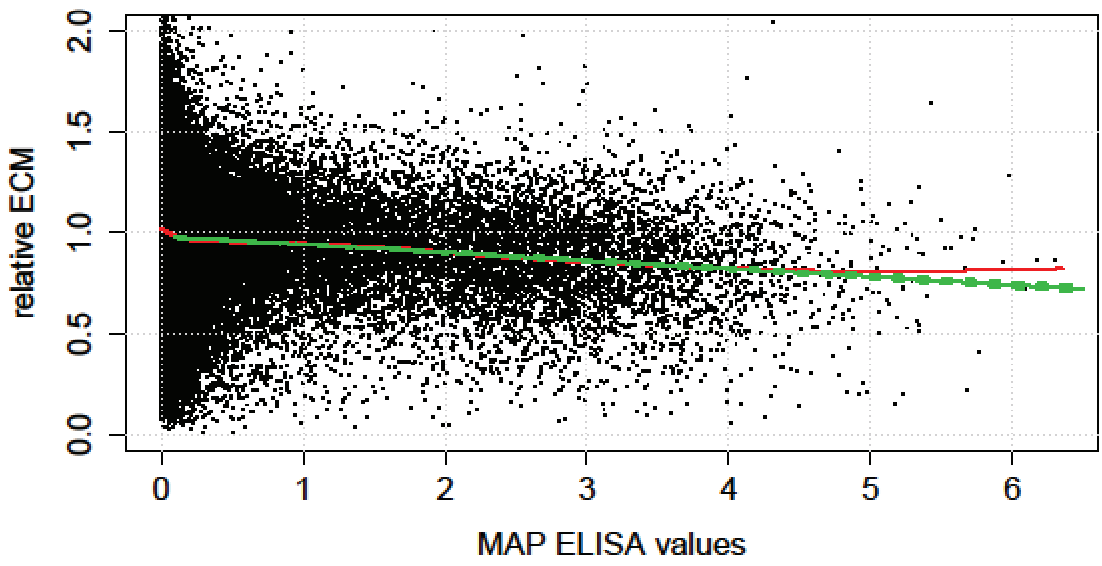

Figure 1: The figure shows the relative energy corrected milk yield (rECM), which is the milk yield relative to the average cow on each farm. The rECM is plotted as a function of the MAP S/P values measured on the same day as the rECM, based on a parity-stratified Wilmink-function. The **red line** is a local polynomial fit. The **green line** is a linear fit in the ELISA range [0.1;6.2], with the dashed green lines representing the 95% confidence interval of the linear fit.

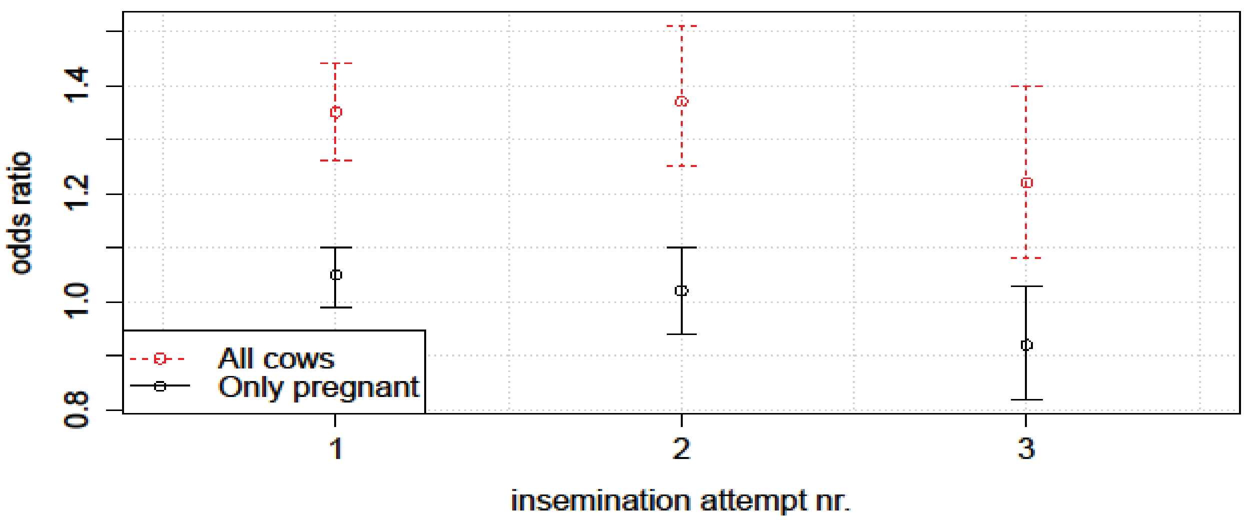

Figure 2: The odds ratio of the conception probability following inseminations 1 to 3 between first and second calving of animals that later tested MAP ELISA positive versus animals that never tested positive. The **red circles** with dashed lines represent the test with all cows, and the **black circles** represent the test with only the cows that achieved proceeding to the following lactation (parity 2), and thereby actually achieved becoming pregnant. Error bars represent the 95% confidence interval of the estimate. The test using all cows suggest that cows that test MAP positive later in life have a ~30% higher chance of a successful insemination. However, when using only cows that do become pregnant, then there is no significant difference between negative and later positive cows.

Milk production of cows prior to the positive MAP ELISA test was tested compared to cows never testing positive. Here the milk production was averaged over the lactation period and the result was that first parity cows that later tested positive produced 2.2% (95% CI: 1.7-2.6%) more milk than cows never testing positive. For the second parity cows, the numbers were 2.7% (95% CI: 1.9-3.3%), and third parity cows produced 3.3% (95% CI: 2.2-4.5) more.

Discussion

The data presented in figure 2 are biased in different ways. When including cows that do not become pregnant (red circle), we include cows that will not live for much longer as they will be culled when milk yield decreases. For this reason the discovery of a MAP infection is less likely as they do not live for long. Therefore, they will count as non-positive cows, and this group will get more cows that never become pregnant, and hence a reduced conception probability compared to later MAP positive cows. When only including cows that eventually become pregnant (black circles) then the excluded cows may have a different distributions of MAP positive animals than the included group, which may draw in either positive or negative direction. So the general problem is that we do not know whether animals are MAP positive, or rather would deliver a MAP positive sample in the future had they survived. However, basing the model design on cows that were already positive would subject the data to a potential decision bias of farmers: that they may be more likely to keep well-functioning MAP positive animals.

That future MAP positive cows produce more milk before testing positive, may be due to high-producing cows being more likely to lose control of the infection, as evidenced by occurrence of antibodies. Another potential reason is that stronger animals may be better to handle the infection. So that they produce more milk is possibly not an effect of the MAP, but because weaker animals succumb earlier to the infection, and the remaining are higher producing animals.

Conclusion

MAP infection prior to being ELISA positive does not appear to negatively influence conception, and may have a positive effect on milk yield for individual cows compared to cows at the same farm. However, positive ELISA values are correlated with a reduction in milk yield, with higher S/P values associated to greater reduction in milk yield.

Data originating from a database should be analysed with caution, because bias may enter in many forms such as bias resulting from farmers' decisions to cull infected but not positive cows, ELISA positive but non-affected (on milk yield or conception) cows etc.

Bibliography

S.S. Nielsen, 2007. Danish control programme on bovine paratuberculosis. Cattle Practice, 31, 150-154.
S.S. Nielsen, N. Toft, H. Okura, 2013. Dynamics of specific anti-*Mycobacterium avium* subsp. *paratuberculosis* antibody response through age. PLoS ONE, 8, e63009.
J.B.M. Wilmink, 1987. Adjustment of test-day milk, fat and protein yield for age, season and stage of lactation. Livesto Pro Sci, 16, 335-348.

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